FIG. 1A


```
                    DMEM        M9      M9 +
                                       NaHCO₃
107 →                                          ← EspC
                                               ← 78kDa
 68 —                                          ← 72kDa
 46 —
                                               ← 40
                                               ← 39
                                                  EspB
 28 —
                                               ← EspA
 19 —
```

FIG. 1B


```
           Membrane  Insoluble        Membrane  Insoluble
           - +   - +  EPEC            - +   - +  EPEC

209 —

107 —
                          ← 90                      ← 90
                          ← Ep85                    ← 78
 68 —
           Anti-PY                    Anti-EPEC78
```

FIG. 2A

Anti-PY -EPEC78

- +   - + CVD206

107 –

Immunoprecipitate

← 90

107 –

Supernatant

Probe-Anti-PY

FIG. 2B

Acidic   **Basic**

209 –

90 → 107 –
        68 –

46 –

Anti-EPEC78

209 –

107 –
90 → 68 –

46 –

Anti-PY

FIG. 3

CVD206

EPEC Supernatant

CVD206

EPEC Supernatant

Alk. Phos.   −   +   −   −   +   −

— 107

← 90

← 78

— 69

— 46

Anti-PY   Anti-EPEC78

FIG. 4

A

E

B   αPY

F   Actin

C   α EPEC78

G   α EPEC78

D   αPY+ α EPEC78

H   Actin+ α EPEC78

FIG. 5

wt
intimin
espA
espB
tir
cfm-14
uninfected

— 107

90 → — 68

Membrane fraction

— 107

90 →
78 → — 68

Insoluble fraction

FIG. 6A

```
CGGCTGCATACCGTTACGTCATAGTAATATAAAGGAACGTGTCAAATTTCTAAATAAAAG    60
     tlr ->
GATATATGTATGCCTATTGGTAACCTTGGTAATAATGTAAATGCCAATCATTTAATTCCC   120
         M  P  I  G  N  L  G  N  N  V  N  G  N  H  L  I  P

CCTGCGCCGCCACTACCTTCACAAACAGACGGCGCGGCACGGGGGAGGAACTGGTCATCTA   180
P  A  P  P  L  P  S  Q  T  D  G  A  A  R  G  G  T  G  H  L

ATTAGCTCTACAGGAGCATTAGGATCTCGTTCATTGTTTTCTCCCCTGAGAAATTCTATG   240
I  S  S  T  G  A  L  G  S  R  S  L  F  S  P  L  R  N  S  M

GCTGATTCTGTCGATTCCAGAGATATTCCAGGACTTCCTACAAACCCATCGAGGCCTTGCT   300
A  D  S  V  D  S  R  D  I  P  G  L  P  T  N  P  S  R  L  A

GCAGCTACATCTGAGACATGCTTGCTTGGAGGATTTGAAGTTCTCCATGATAAGGGGCCA   360
A  A  T  S  E  T  C  L  L  G  G  F  E  V  L  H  D  K  G  P

CTTGATATTCTCAATACGCAAATTGGACCCTCTGCATTTCGTGTTGAAGTGCAGGCAGAT   420
L  D  I  L  N  T  Q  I  G  P  S  A  F  R  V  E  V  Q  A  D

GGTACTCATGCCGCTATTGGAGAAAAAAATGGTTTGGAGGTTAGCGTTACATTAAGTCCT   480
G  T  H  A  A  I  G  E  K  N  G  L  E  V  S  N  T  L  S  P

CAAGAATGGAGCAGCTTGCAATCTATTGATACTGAGGGTAAAAACAGATTTGTTTTTACC   540
Q  E  W  S  S  L  Q  S  I  D  T  E  G  K  N  R  F  V  F  T

GGGGGACGTGGCGGTAGTGGGCATCCGATGGTCACTGTCGCATCAGATATCGCGGAAGCT   600
G  G  R  G  G  S  G  H  P  M  V  T  V  A  S  D  I  A  E  A

CGTACGAAATACTGGCCAAATTAGACCCAGACAATCATGGAGGACGTCAACCCAAGGAC   660
R  T  K  I  L  A  K  L  D  P  D  N  H  G  G  R  Q  P  K  D

GTTGATACGCGTTCTGTTGGTGTTGGCAGCGCCTTCGGGAATAGATGATGGCGTTGTTAGC   720
V  D  T  R  S  V  G  V  G  S  A  S  G  I  D  D  G  V  V  S

GAAACCCATACTTCAACAACAAATTGGAGCGTTCGCTCAGATCCTAAATTCTGGGTTTCT   780
E  T  H  T  S  T  T  N  S  S  V  R  S  D  P  K  P  W  V  S

GTCGGCGCAATTGCTGCTGGTTTAGCGGGACTGGCGGCAACTGGTATTGCACAGGCGTTG   840
V  G  A  I  A  A  G  L  A  G  L  A  A  T  G  I  A  Q  A  L

GCTTTGACACCGGAACCGATGATCCTACAACCACCGATCCTGATCAGGCCGCAAATGCT   900
A  L  T  P  E  P  D  D  P  T  T  T  D  P  D  Q  A  A  N  A

GCAGAAAGTGCAACAAAAGATCAGTTAACGCAAGAAGCATTCAAGAACCCTGAGAACCAG   960
A  E  S  A  T  K  D  Q  L  T  Q  E  A  F  K  N  P  E  N  Q

AAAGTTAACATCGATGCGAACggaaatgctattccgtctgggggaattaaaagatgatatt  1020
K  V  N  I  D  A  N  G  N  A  I  P  S  G  E  L  K  D  D  I

gttgagcaaatagcacaacaagctaaagaggctggtgaggtggccagacagcaggctgtt  1080
V  E  Q  I  A  Q  Q  A  K  E  A  G  E  V  A  R  Q  Q  A  V

gaaagcaatgcacaggcgcagcagcgatatgaggatcagcatgccagacgtcaggaggaa  1140
E  S  N  A  Q  A  Q  Q  R  ▮  E  D  Q  H  A  R  R  Q  E  E

ttacagctttcatcgggtattggttacggcctcagcagtgcattgattgttgctggggga  1200
L  Q  L  S  S  G  I  G  ▮  G  L  S  S  A  L  I  V  A  G  G

attggtgctggtgtaacgactgcgctccatagacgaaatcagccggcagaacagacaact  1260
I  G  A  G  V  T  T  A  L  H  R  R  N  Q  P  A  E  Q  T  T

actacaacaacacatacggtagtgcagcaacagaccggagggggataccccagcacaaggtg  1320
T  T  T  H  T  V  V  Q  Q  Q  T  G  G  I  P  Q  H  K  V

gcactgatgccacaagagcgaagacgcttctctgatagacgtgattcgcaggggagtgtt  1380
A  L  M  P  Q  E  R  R  R  F  S  D  R  R  D  S  Q  G  S  V

gcatcgacacactggtcagattcctctagcgaagtggttaatccatatgctgaagttggg  1440
A  S  T  H  W  S  D  S  S  S  E  V  V  N  P  ▮  A  E  V  G

ggggctcggaatagtctatcggctcatcagccagaagagcatatttatgatgaggtcgct  1500
G  A  R  N  S  L  S  A  H  Q  P  E  E  H  I  ▮  D  E  V  A

gcagatcctggttatagcgttattcagaatttttcaggggagcggcccagttaccggaagg  1560
A  D  P  G  ▮  S  V  I  Q  N  F  S  G  S  G  P  V  T  G  R

ttaataggaactccagggcaaggtatccaaagtacttatgcgcttctggcaaacagcggc  1620
L  I  G  T  P  G  Q  G  I  Q  S  T  ▮  A  L  L  A  N  S  G

ggattgcgtttaggtatggggaggattaacgagtggtggcgagacggcagtaagttctgta  1680
G  L  R  L  G  M  G  G  L  T  S  G  G  E  T  A  V  S  S  V

aatgccgcaccaacgccgggaccagtacgtttcgtttaaatatatctgtgagtatttagt  1740
N  A  A  P  T  P  G  P  V  R  F  V  •

tgaggttggggtggggtgggggggcgttttactagcgttaatgtttcagagaacaacgtt  1800
                              orfU ->
gcagcatgggtaactcttgaacttctgttattataatcaattaagagaaattataatgtc  1860
                                                       M  S

atcaagatctgaactttttattagataggtttgcggaaaaaattggtgttggatctatttc  1920
S  R  S  E  L  L  L  D  R  F  A  E  K  I  G  V  G  S  I  S
```
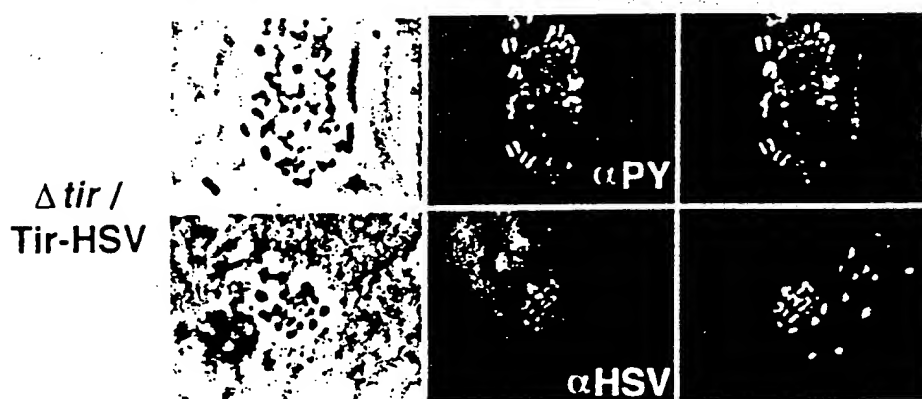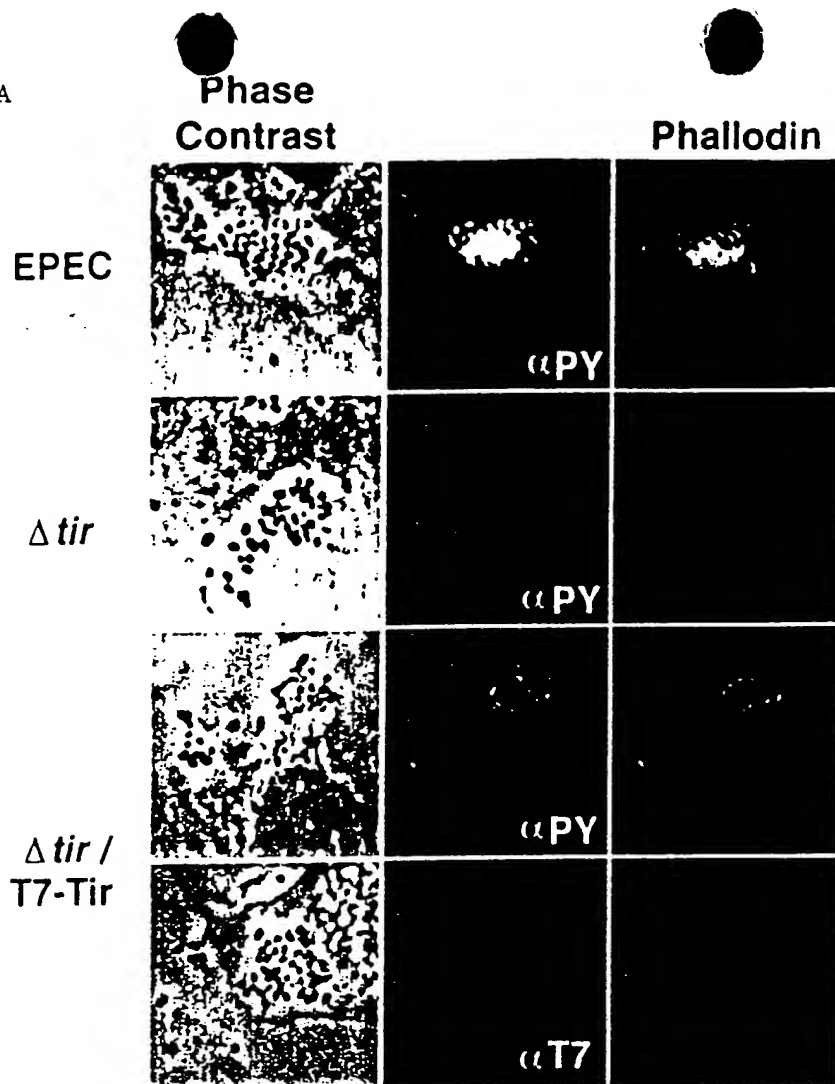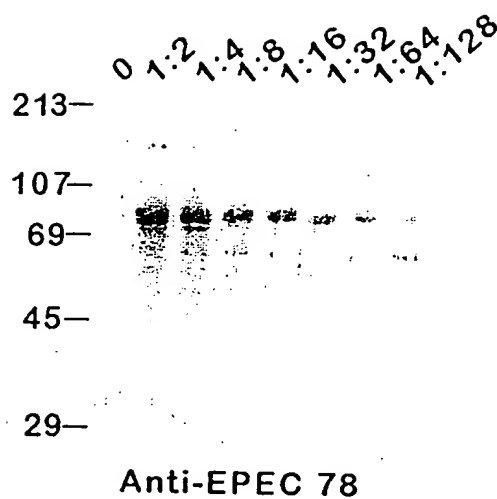
FIG. 6B

EcoRI   BamHI   EcoRV   ClaI   BglII   EcoRI

tir   *orfU*   *eaeA*

EcoRI   BamHI   ClaI   BglII   EcoRI

Sal I

Δ *tir*   *orfU*   *eaeA*

FIG. 7A

Phase
Contrast   Phallodin

EPEC

αPY

Δ tir

αPY

Δ tir /
T7-Tir

αPY

αT7

Δ tir /
Tir-HSV

αPY

αHSV

FIG. 7B

1 2 1 2 1 2

T7-Tir
Tir   Membrane fraction

T7-Tir   T7 immunoprecipitate

Anti: PY   T7   EPEC78

FIG. 8A

213—

107—
69—

45—

29—

0 1:2 1:4 1:8 1:16 1:32 1:64 1:128

T7-Intimin Overlay

213—

107—
69—

45—

29—

0 1:2 1:4 1:8 1:16 1:32 1:64 1:128
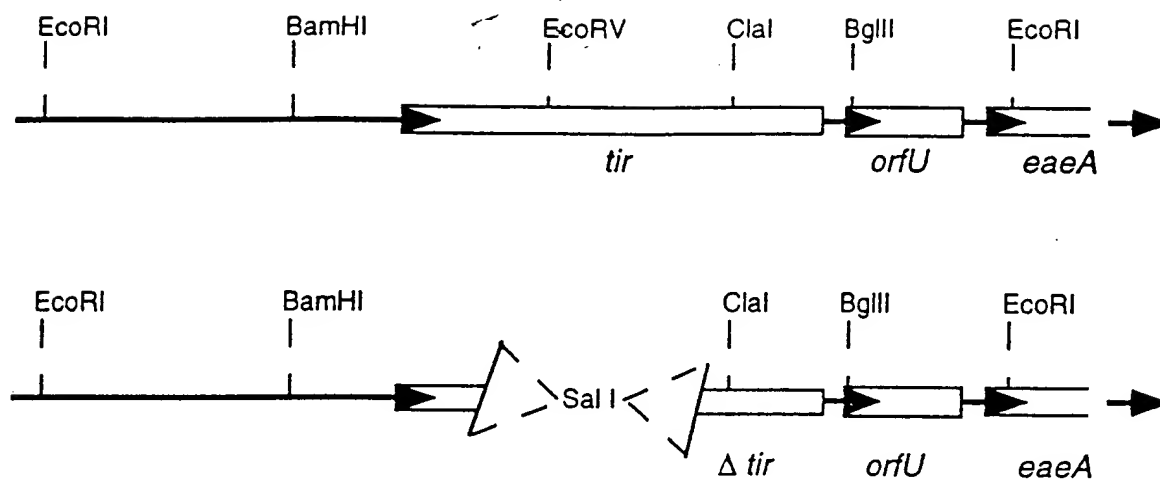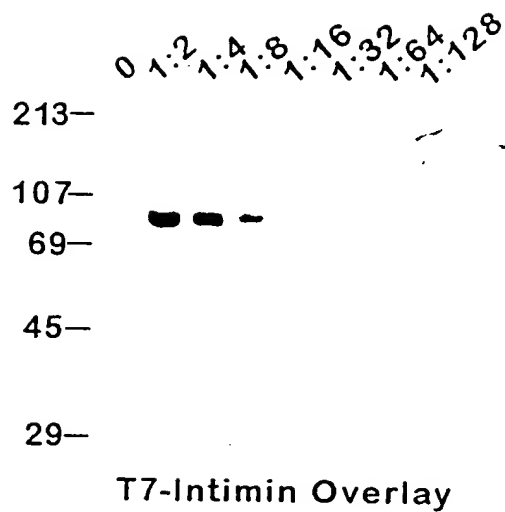
Anti-EPEC 78

FIG. 8B
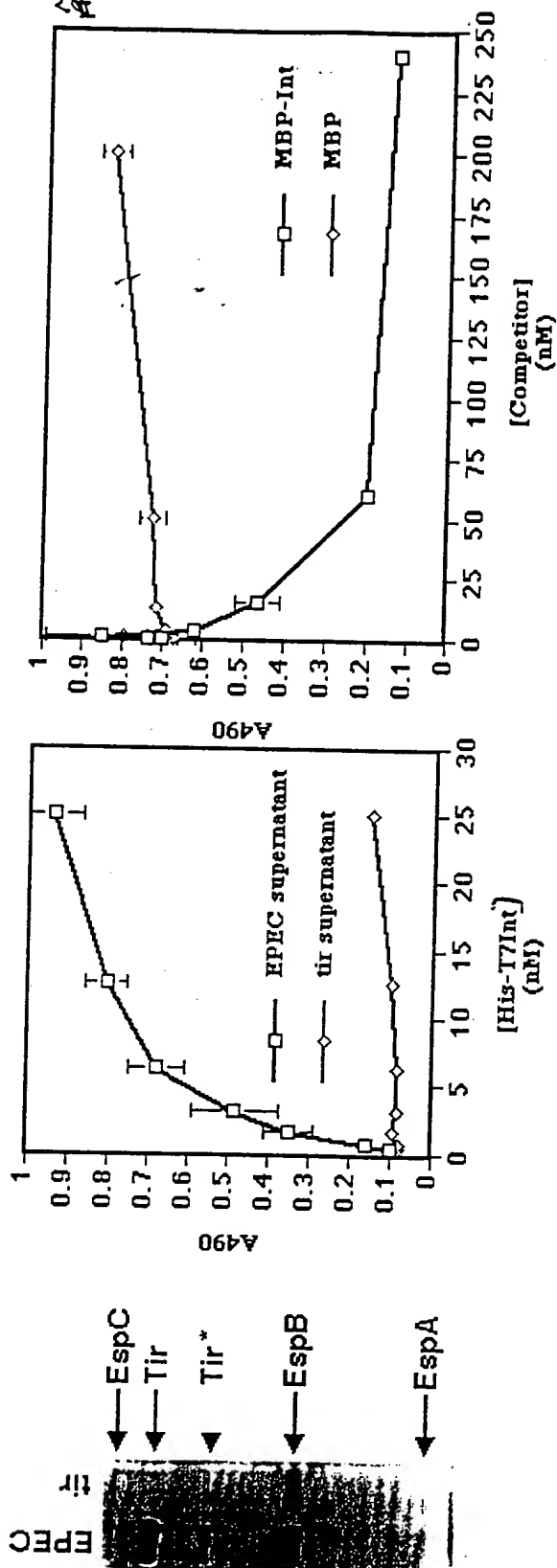
FIG. 9

```
EPEC     1   MPLGNLQNNVNGNHLIPPAPPLPSQTDGAARGQTGHISS...
EHEC     1   MPLGNLQHNPNVNNSIPPAPPLPSGTDGAG--GRQQELNS...
RDEC-1   1   ...........................................

EPEC    43   ALGSRSLFSPLRNSMADSVDSR--DIPQLPTNRSILAAATSE
EHEC    41   PLGSRALPTPVRNSMADSGDNRASDVRGLPVNBMRLASS--
RDEC-1   1   .........NSVADAAAESRASDIPQLPTNRLRFASS-E

EPEC    83   TCLLGQRPVLHDKGPLDIENTQLGPSARRVEVGADSRAALG
EHEC    81   ITNDGPEVLHDHGPLDTENRQTGSSVFRVPTQEDGKTIAVE
RDEC-1   29  VSEHGALEVLHDKGGLDTLNSAIGSSLFRVETRDDGSHVALG

EPEC   125   EKNGEEVSVTFSPQEWSSLQSIDTEGKNRFVETQGRLGSHIP
EHEC   123   QRNGVETSYVLSDQEYARLQSLDFEGKDKFVCTGCRGGAULSA
RDEC-1   71  QKNGLETTVVLSEQEFSSLQSLDFEGKNKFVETGGRSSPEE

EPEC   167   MVIYASDIAFARTKTLAKIDRDNHGGRQPKDVDRRSEVGSA
EHEC   165   MVIYASDIFPARQRLDELLPEKGTG-ESKGAGESKGESLRE
RDEC-1   113  MVIYASDIAPARQRIIDKLPEK.........DIKETKPGD

EPEC   209   SGIDDGVVSELTSTTNSSVRSDPKFWVSVGATAAGLAGGAA
EHEC   206   SKSGAENTTETQTSTSTSSLRSDPKTWGALGTVATPARGLLA
RDEC-1   145  PKSGEGKIIEIHTSVTSSLRADPKLWLSGQTTAAGLAGGMAS

EPEC   251   TGIAQAIALTPEPDDIQTTTDPDQAANAAESAIKDIFTQLGK
EHEC   243   IGIVQALSLTPEPDSPFTTDPDAAASATETARDQLLLLGEG
RDEC-1   137  TGIAQAVALTPEPDDPITTDPDAAANTAEAKAKDQLFLLG

EPEC   293   XPENQKVNIDANGNAIPSGELKDDIVEQIAQQAKEAGLVLHG
EHEC   290   NPDNQKVNIDELGNAIPSGVEKDDVVANLEEQAKAAGHEKE
RDEC-1   229  NPDNQKVNIDENGVAIPSGELKDDVVAQIAEQAKAAGSEQLR

EPEC   335   QSVESNAQAQQRLREDQHARRQRELQLSSGIGHGESSAEVAG
EHEC   332   QATENNAQAQKKLDEQQAKRQEELKVSSCAGYGLSQALLSS
RDEC-1   271  EAIESNSQAQQKYDEQHAKREQEMSSSQVGYGIRGALLGG

EPEC   377   GIGAGVTTALHRNQRAEQTTTTTI......HLVVQQQTLGG
EHEC   374   GIGVAVTAALHRKNQPVEQTTTTTTTTTTSARTVENKPANN
RDEC-1   313  GIGAGVTAALHRKNQPAEQTITTRT......VVDNQFTNN

EPEC   412   IPQHKVALMPQERRRFSDRRDSQGSVASHWSDESS-EMV
EHEC   416   TPAGGNVDTPGSEDTMESRRSSMASTSSTFFDRSQILQX
RDEC-1   347  ASAQQNTDTSGPEESPASRRNSNASLASNGSDRTCESSEXE

                                    473
EPEC   453   YAEVGGARNSLSAHQPEEHIYDEVAADPG...YCVTONES.E
EHEC   453   YADVKTSLHDSQVPTSNSNTSVQNMGNTDSVYSLTSSPPRD
RDEC-1   339  YADVGMPRNDSLARISEEPIYDEVAADPN...YSXTGSS.E

EPEC   491   SGPVTGRLIQTPQQQCIQSTYALLANSGGLRLGMGGLTSGLT
EHEC   500   TTDNGARLLGNPSAQTQSTYARLALSGGIRHDMGGLTTSN
RDEC-1   427  NSRVTQRLVGTPQQQIQSTYALLASSGGLRLGMGGLTTGT

EPEC   533   AVSSVNAAPTPQPVRFV
EHEC   542   AVNTSNNPPAFGSHRPV
RDEC-1   469  AYSTANAARLPGPAREV
```